

# **AMENDMENTS TO THE CLAIMS**

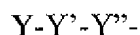
This listing of claims will replace all prior versions, and listings, of claims in the application:

1-90 (Cancelled).

91. (New) A method of obtaining covalent or catalytic antibodies from organisms with autoimmune or alloimmune disease, an organism without known disease or transgenic mice expressing human antibody genes, comprising isolating the antibodies therefrom using a covalently reactive polypeptide antigen analogue (pCRA) of formula (I)



wherein L is a ligand that binds noncovalently to a nucleophilic receptor Nu and E is an electrophilic group conjugated to a side chain functional group of L having the formula



wherein

Y'' is an atom, covalent bond or linker,

Y' is an atom, bond or chemical group that connects Y and L or Y'',

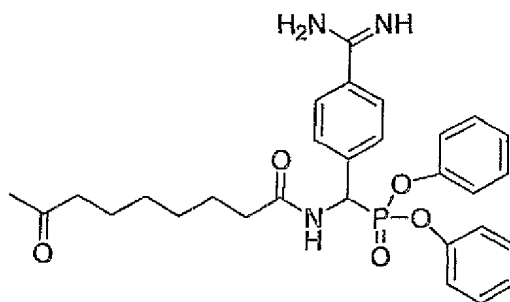
Y is a covalently reactive electrophilic group that reacts specifically with an antibody that binds to L.

92. (New) A method according to claim 91, in which any one of Y, Y', or Y'' contains a water-binding group as a terminal or internal component (pCRAW).

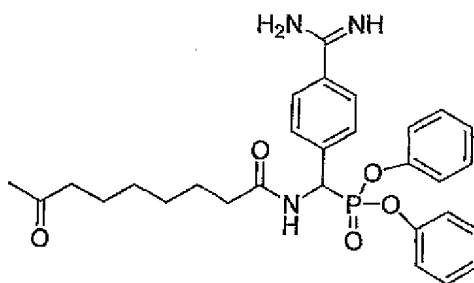
93. (New) A method of obtaining covalent or catalytic antibodies from organisms with an autoimmune or alloimmune disease, an organism without known disease or transgenic mice expressing human antibody genes, in which the organism is immunized using a covalently reactive polypeptide antigen analogue according to claim 92.

94. (New) The method according to claim 91, in which Y", Y' or Y contains a water binding group as a terminal or internal component that is composed of a site that binds a zinc, copper, nickel, cobalt, calcium or magnesium ion which chelates one or more water molecules in which the metal binding site is  $-(\text{His})_n$  wherein  $n=2$  or more,  $-\text{Cys-X-Cys-Cys-}$  or  $-\text{Cys-X-Cys-}$  peptide regions wherein X is an amino acid residue, ethylene diamine tetraacetic acid or diaminomethyl pyridine.

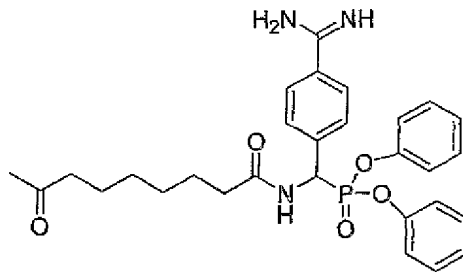
95 (New) The method according to claim 91, wherein L represents an antigenic determinant or polypeptide from a microbial protein, a human, animal or plant protein, an antigen that is over-expressed on cancer cells, Factor VIII, epidermal growth factor receptor, CD4, beta-amyloid peptide 1-40 or beta-amyloid peptide 1-42 or the epidermal growth factor receptor derivatized at one or more Lys side chain amino group with



or, is vasoactive intestinal peptide derivatized at the Lys20 side chain with



or, is the HIV-I protein gp120 derivatized at Lys side chain amino group at a density of 23 moles/mole protein with



96. (New) The method according to claim 91, wherein the antibodies are:  
 polyclonal antibodies identified in the serum of the organism by steps comprising:
- a) screening and selection for covalent antibodies by binding to the antigenic pCRA or a polypeptide, wherein the binding is optionally determined in sodium dodecyl sulfate; and
  - b) screening and selection for catalytic hydrolysis of a suitable substrate, or monoclonal antibodies or antibody fragments obtained from lymphocytes of the organism by steps comprising:
    - i) preparing from the lymphocytes a library of hybridoma cell lines, virus-transformed cell lines or immunoglobulin fragment genes cloned in and expressed from a vector, wherein the lymphocytes are optionally contacted with the pCRA or a polypeptide, and lymphocytes that bind the pCRA or polypeptide are separated from lymphocytes that do not bind the pCRA or polypeptide;
    - ii) screening and selecting for covalent antibodies or antibody fragments by binding to the antigenic pCRA or a polypeptide;
    - iii) screening and selecting for catalytic hydrolysis of a suitable substrate; and
    - iv) purifying the antibodies or the antibody fragments.

97. (New) The method according to claim 96, wherein the antibody fragments are single chain Fv fragments containing the VL and VH domains or light chains expressing covalent or catalytic activity isolated by steps comprising:

- a) preparing the immunoglobulin VL cDNA, VH cDNA and light chain cDNA by reverse-transcriptase polymerase chain reaction using as template the RNA from lymphocytes;
  - b) cloning the VL and VH cDNA in a form enabling their expression as single chain Fv fragments expressed on the surface of a display vector;
  - c) cloning the light chain cDNA in a vector in a form enabling their expression as light chains expressed on the surface of a display vector;
  - d) contacting the vector particles with immobilized pCRA or polypeptide, removing unbound vector particles by washing, and expressing the Fv cDNA or light chain cDNA from the pCRA-bound vector particles in soluble form in prokaryotic or eukaryotic cells;
  - e) screening the soluble Fv or light chain constructs for covalent antigen binding activity;
- and
- f) screening the soluble Fv or light chain constructs for catalytic activity.

98. (New) The method according to claim 97, further comprising improving the covalent or catalytic activity of the antibody fragments by the further steps of:

- a) introducing mutations in the VL or VH domains or both;
- b) displaying the resultant antibody fragments on the surface of a display vector;
- c) contacting the vector particles with the pCRA or polypeptide and removing the unbound vector particles;
- d) expressing the antibody fragments in soluble form in prokaryotic or eukaryotic cells;
- e) screening the antibody fragments for covalent antigen binding activity;

f) screening the antibody fragments for catalytic activity.

99. (New) The method according to claim 97 further comprising preparing full-length IgG, IgA, IgM, IgD or IgE antibodies from the Fv fragments by steps comprising:

a) insertion of the VL and VH domain DNA at the 5' side of Ig constant domains contained in an expression vector by nucleic acid digestion and ligation procedures;

b) growth of the vectors in a prokaryotic or eukaryotic host cell, extraction of the full-length antibodies from the culture medium or the cellular contents and purification of said antibodies.

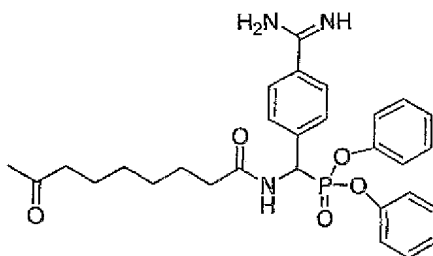
100. (New) The method according to claim 97 further comprising preparing full length IgG, IgA, IgM, IgD or IgE antibodies from the light chain fragments by steps comprising:

a) insertion of the cDNA encoding the VL domain with covalent or catalytic activity into an expression vector containing the constant domain of the light chain by nucleic acid digestion and ligation procedures;

b) insertion of the cDNA encoding the VH domain with noncovalent antigen binding activity into an expression vector containing the constant domains of the heavy chain by nucleic acid digestion and ligation procedures;

c) growth of the vectors in a prokaryotic or eukaryotic host cell, extraction of the full-length antibodies from the culture medium or the cellular contents and purification of said antibodies.

101. (New) The method according to claim 96, wherein the monoclonal antibodies bind gp120 or the gp120-CRA which is derivatized at the Lys side chain amino group at a density of 23 moles/mole protein with:



and wherein is resistant to dissociation with 2% SDS.

102. (New) The method according to claim 96 further comprising preparing from the covalent or catalytic monoclonal antibodies or antibody fragments a therapeutic or prophylactic composition that inactivates or neutralizes one or more antigens associated with a medical disorder or condition in an organism.

103. (New) The method according to claim 91, further comprising preparing from the pCRA an antigenic composition effective to inhibit the action of an antibody in an individual with a medical disorder or condition, wherein one or more antigenic determinant are bound irreversibly by the antibody.

104. (New) The method according to claim 91 further comprising preparing from the pCRA an immunogenic composition that stimulates the production by an organism of therapeutic or prophylactic antibodies having covalent or catalytic activity specific for an antigen associated with a medical condition or disorder in the organism.

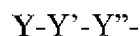
105. (New) The method according to claim 102, wherein the medical disorder is an infectious disease, HIV, hepatitis, Alzheimer' disease, a cancer, an autoimmune disease, a lymphoproliferative disorder, a blood proliferative disorder or a genetic defect.

106. (New) A water-binding, covalently reactive polypeptide antigen analogue (pCRAW) of formula (1):

L-E

(I)

wherein L is a ligand that binds noncovalently to a nucleophilic receptor Nu and E is an electrophilic group conjugated to a side chain functional group of L having the formula



wherein

Y'' is an atom, covalent bond or linker,

Y' is an atom, bond or chemical group that connects Y and L or Y'',

Y is a covalently reactive electrophilic group that reacts specifically with an antibody that binds to L,

Y'', Y' or Y contains a water-binding group as a terminal or internal component with a site that binds a zinc, copper, nickel, cobalt, calcium or magnesium ion which chelates one or more water molecules in which the metal binding site is -(His)<sub>n</sub>- where n=2 or more, or Cys-X-Cys-Cys or -Cys-X-Cys- wherein X is an amino acid residue, ethylene diamine tetraacetic acid or diaminomethyl pyridine.

107. (New) A monoclonal IgG antibody obtained with the method of claim 91.